Parathyroid hormone-related peptide stimulates intestinal strontium absorption in Camels (*Camelus dromedarius*)

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Abstract

The present work was undertaken to evaluate the stimulatory effect of Parathyroid hormone-related peptide (PTHrP) on the intestinal calcium (Ca) absorption using stable strontium (Sr) as a surgoate marker in 10 Camels. The animals were randomly divided into two groups of five animals. Just after an oral Sr load (4.1 mmol of $SrCl_2$), the first and the second (control) groups received either an i.v infusion of synthetic human PTHrP or solvent alone respectively. PTHrP induced a significant rise in levels of plasma Sr (52.8 ± 4.6 µmol/L VS 41.7 ± 4.5 µmol/L, $P < 0.05$, at the second hour after oral Sr load) and urinay Pi excretion comparatively with animal controls. Plasma Sr levels remained very higher in treated animals than those measured in controls until the fifth hour of experimentation (52 ± 4.7 µmol/L VS 42.1 ± 4.5 µmol/L, $P < 0.05$). PTHrP did not induce significant variation on Ca or Sr renal excretion. Our results seem to show that PTHrP may play an important role on modulation of intestinal Ca absorption in camels.

Keywords: Calcium / strontium / intestinal absorption / PYHrP / dromedary.

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1. Introduction

It's commonly accepted that intestinal Ca^{2+} absorption is a crucial control system in the regulation of Ca_2^+ homeostasis, because it facilitates the entry of dietary Ca^{2+} into the extracellular compartment by two distinct mechanisms including passive (paracellular) and active (transcellular) transport (Hoenderop et al., 2005). The vitamin D metabolite 1,25-dihydroxyvitamin D3 [1,25-(OH)2D3], parathyroid hormone (PTH) and PTH-related peptide (PTHrP) among others, are prominent hormones controlling the Ca^{2+} balance (Hoenderop et al., 2005). Early studies demonstrated that in domestic ruminants, PTHrP which is produced during gestation by fetal parathyroid glands and placenta (Rodda et al., 1988) and during lactation by mammary glands (Thiede, 1989) can play an important role in the regulation of phosphocalcic metabolism. In previous experimental studies which has been carried out in Morocco in camels, we have reported in part that the

physiological levels of PTHrP in colostrum induced a postprandial hypercalcemia (El Khasmi et al., 2000) and in part, that Sr absorption test was reproductible and might be used to explore intestinal Ca absorption (El Khasmi et al., 2003). So, in the work reported here, we have evaluated the effect of PTHrP on intestinal Ca absorption by using stable Sr as a surgoate marker in camels.

2. Materials and methods

This study was carried out in the south of Morocco (Laâyoune areas) on 4-6-month-old female camels (*Camelus dromedarius*) weighing 140 ± 20 Kg (Mean ± SEM). The animals were healthy during experimentation and were fed a daily ration of hay and barley, providing a daily intake of 30 g Ca and 10 g P. They were watered daily. The camels were randomly divided into two groups of five animals and orally received within 1 min, 360 ml of a solution containing 4.1 mmol of $SrCl_2$ (Sigma, Aldrich). At the end of oral Sr load, the animals from the first group were infused through an indwelling catheter into the left jugular vein, with synthetic human PTHrP (1-34) fragment (from Bissendorf Biochemicals GmBH, Hannover, Germany). The dose used (4 nmol/Kg body wt) was dissolved in CH3COOH 0.1 M and diluted with sterile 0.9% NaCl containing 0.1% bovine serum albumin. Half the dose was given i.v as a quick bolus injection at 9 a.m., the second half through a 1 hr i.v. infusion starting immediately after injection. Those from the second (control) group received in the same way the same volume (25 ml) of solvent alone. Urine excretion of each camel was measured during the 6 hr following hormonal treatments. Urine was collected using plastic bags designed for the urinary tract of female camels (Bengoumi et al., 1993). For each animal, the volume of urine collected during a period was measured, and a 10 ml sample was collected and frozen at -20°C until analysis. Blood samples were collected by puncture from the right jugular vein at the beginning of the experimentation (o time), and 1 hr, 2 hr, 3 hr, 4 hr and 6 hr thereafter. After centrifugation at 1500 g for 10 min, plasma was collected and frozen. In thawed plasma and urine samples, Ca was measured by atomic absorption spectrophotometry (Perkin Elmer 560). P was measured by colorimetry. The Sr levels were measured by atomic absorption spectrophotometry at 460.7 nm with use of an acetylene-air flame in 10-fold-diluted plasma and 50-fold-diluted urine with 20 g/L lanthanum and 100 ml/L hydrochloric acid as the diluent. Results are presented as mean ± SEM. The Mann-Whitney *U* test was used for comparison between groups. Within each group, values measured during or after treatment were compared with those measured before treatment using one-way analysis of variance.

3. Results

PTHrP increased plasma Sr and calcemia and decreased phosphatemia (Fig. 1). At the second hour after oral Sr load, plasma Sr (µmol/L) in camels given PTHrP is significantly very higher than that measured at the same time in controls (52.8 ± 4.6 VS 41.7 ± 4.5, $P < 0.05$). In PTHrP treated animals, plasma Sr levels (µmol/L) remained very higher comparatively with controls until the fifth hour of experimentation (52 ± 4.7 VS 42.1 ± 4.5, $P < 0.05$) (Fig. 1). In camels given PTHrP, plasma Ca (mmol/) increased from 2.30 ± 0.04 just before injection to 2.90 ± 0.05 ($P < 0.05$) 2 hr after the end of the infusion. Simultaneously, in these animals, plasma P (mmol/L) decreased from 1.87 ± 0.02 to 1.45 ± 0.04 ($P < 0,05$) (Fig. 1). Urine volume (26.7 ± 3.4 ml/h) and urinary Sr and Ca concentrations (mmol/L) (0.47 ±

0.04 and 0.46 ± 0.01 respectively) measured in controls were never significantly different from that measured in treated animals (Fig. 2). However, urinary P concentration (mmol/L) (0.22 ± 0.03 in controls) was increased by PTHrP (0.6 ± 0.07, $P < 0.05$) (Fig. 2).

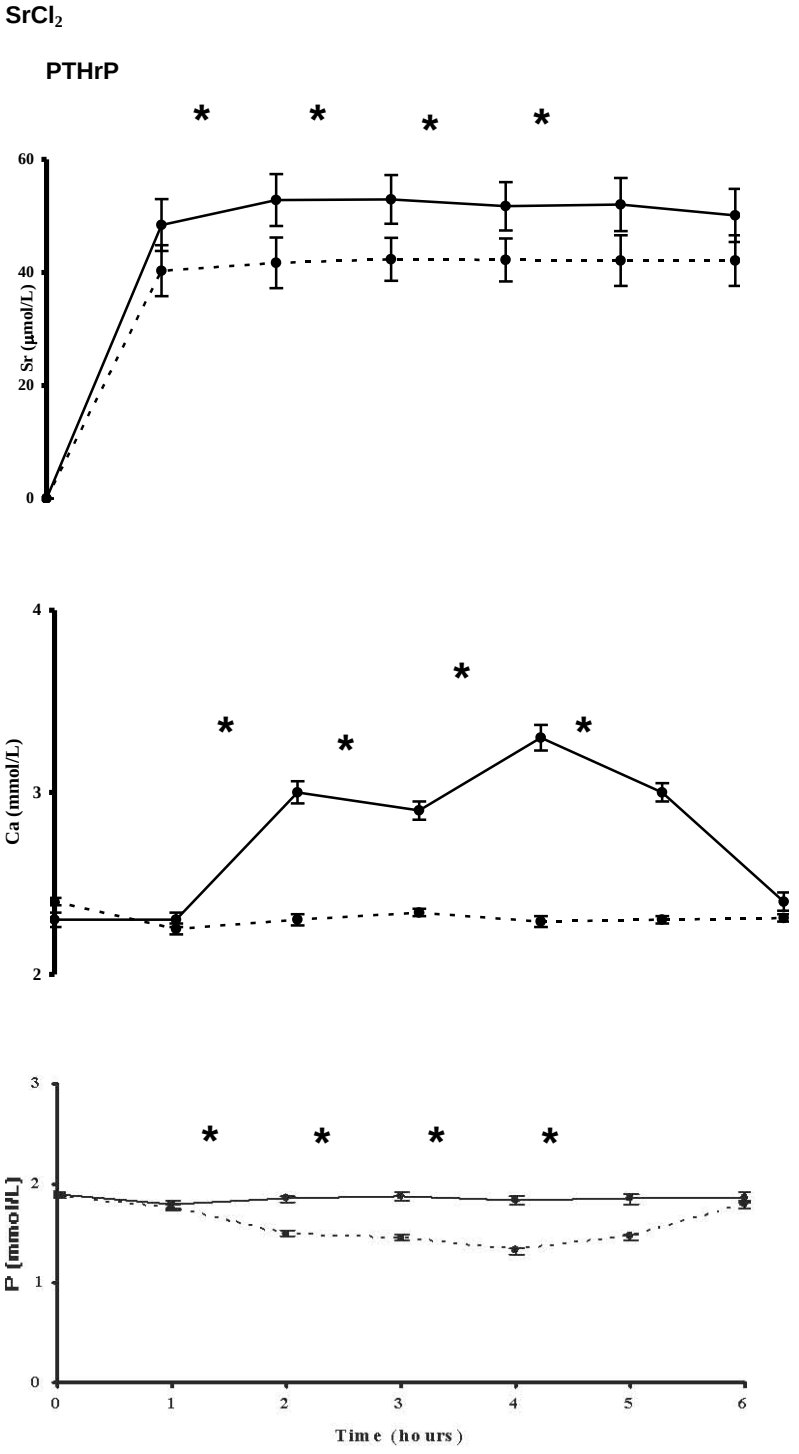

Figure 1. Urine volume and urinary concentration of Sr (white bars), Ca (stripped bars) and P (black bars) measured during the 6 hr-period following PTHrP (1-34) injection just after an oral load of 4.1 mmol of $SrCl_2$ in five camels (means ± SEM; * $P < 0.05$, comparison with controls).

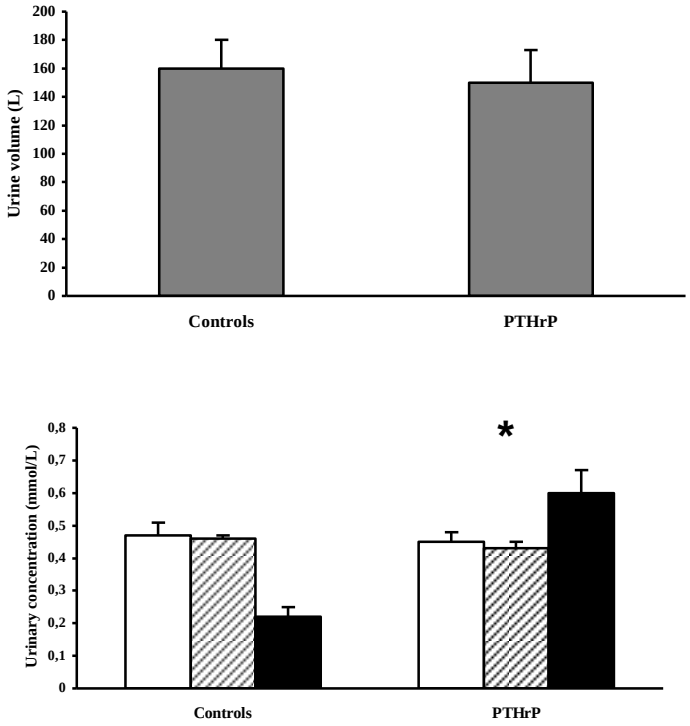

Figure 2. Urine volume and urinary concentration of Sr (white bars), Ca (stripped bars) and P (black bars) measured during the 6 hr-period following PTHrP (1-34) injection just after an oral load of 4.1 mmol of $SrCl_2$ in five camels (means ± SEM; * $P < 0.05$, comparison with controls).

4. Discussion

In this experimentation, we have evaluated the effect of PTHrP on intestinal Ca absorption by using stable Sr which because of its good reproducibility, could be used as a surgoate marker in camels (El Khasmi et al., 2003). In this species, preliminary studies suggested that PTHrP appeared able to contribute to this function (El Khasmi et al., 2005). In fact, it is largely accepted that transport of Sr ions through enteral and renal tubular cells is mediated by the same membrane carriers as used for Ca, and a highly significant correlation has been observed between S rand Ca absorption (Hart and Spencer, 1967; Milson et al., 1987). Furthermore, a comparison of Sr test with the single isotope radio-Ca absorption test in the same group of patients showed a close correlation between the fractional absorption rates of the two elements (Reid et al., 1986). Therefore, the oral administration of stable Sr is considered suitable for assessing Ca absorption and excretion in clinical practice in man (Vezzoli et al., 1995), rat (Corradino et al., 1971) and domestic ruminants (Comar and Wasserman, 1964; Gibbons et al., 1972; Wadhawa and Care, 2000).

The available data show that intestinal transfer of Sr during the first hour depended more on duodenal and jejunal absorption efficiency (Leeuwenkamp et al., 1990; Sips et al., 1994). According to Dumont et al., (1960), Sr transfer is passive only and regulated by humoral factors. Whereas, others findings suggested that Sr transport across the intestinal wall is active (Hendrix et al.,

1963; Wasserman, 1988) and vitamin D dependant (Sips et al., 1997).

In camels given PTHrP, high plasma levels of Sr (figure 1), strongly indicated a high Sr intestinal absorption. In newborn camels, we have demonstrated in previous study that PTHrP stimulated intestinal absorption of D-Xylose (El Khasmi et al., 2000). These stimulatory effects could be mediated by activation of intestinal cell differentiation and modulation of gastrointestinal motility (Budayr et al., 1989; Barlet, 1993). In addition, PTHrP was revealed able to stimulate Xylose intestinal absorption in rat and pig by induction a relaxation of gastrointestinal tract (Mok et al., 1989), Ca intestinal absorption in chiken (Zhou et al., 1992) and ruminal absorption of Ca and phosphorus in sheep (Dua et al., 1994). The PTHrP is an important paracrine/autocrine regulator of proliferation, apoptosis, and differentiation in several normal cell types (Massfelder et al., 1997). In the same way of parathyroid hormone (PTH), PTHrP binds equivalently to a common G protein coupled receptor, the PTH/PTHrP receptor (Abou-Samra et al., 1992), which mediates the endocrine actions of PTH on mineral ion homeostasis and the multiple actions of PTHrP on different tissues in adult and fetus (Massfelder et al., 1996).

In our animals, the PTHrP can mediate the Ca transfer across intestinal epithelia by distinct processes which include passive paracellular (direct exchange between two compartments) and active transcellular (transport across at least two plasma membrane barriers) pathways. According to Hoenderop et al., (2005), transcellular Ca absorption is active and located largely in the duodenum and upper jejunum, whereas paracellular Ca absorption is passive and occurs throughout the entire length of the intestine in rat.

In camels, the PTHrP may influence the Ca movement across intestinal epithelia and may be prominent hormone controlling the Ca balance.

5. References

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